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ON THE OCCURRENCE OF AN ISOLATED ANTIBODY IN THE CEREBROSPINAL FLUID.*

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In the work by Dr. Carlson and myself on the distribution of antibodies in the body fluids of dogs,[1] we found that in dogs, injected with rat blood, opsonin for rat corpuscles appeared in the cerebrospinal fluid, which, however, did not contain a trace of agglutinin. In the serum of the blood and the lymph, both these antibodies described parallel curves. The curve of the opsonin concentration in the cerebrospinal fluid described exactly the same course as in the blood and lymph, but remained much lower. In Chart 1 are reproduced composite curves showing these relations and obtained from estimations on dogs killed at varying intervals after the intravenous injection of one cubic centimeter of 10 per cent suspension of rat blood per kilo of the weight of dog.

In dogs injected intravenously with goat blood we did not find any agglutinin in the cerebrospinal fluid. During the period of highest antibody content in the blood and lymph, however, the fluid contained traces of lysin and opsonin.

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[1] *Jour. Infect. Dis.*, 1910, 7, p. 319.

The fact that opsonin and not agglutinin appeared in the cerebrospinal fluid of dogs injected with rat blood was interesting because it seemed to point to the separate entity of these antibodies. There was a possibility, however, that agglutination and opsonification might be caused by one substance, the agglutinative action of which was suppressed by the influence of the cerebrospinal fluid. Consequently, on account of the theoretical importance of the question of the separate entities of the bodies concerned in the various forms of antibody action, its seemed desirable to study the antibody

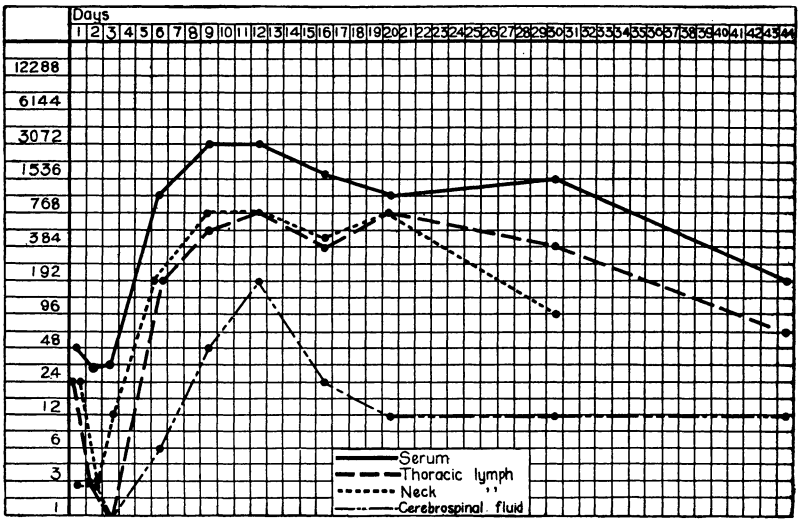

CHART 1.—Specific opsonin in blood, lymph, and cerebrospinal fluid of dogs injected with rat blood.

content of the cerebrospinal fluid of dogs a little more closely. In this article is given a brief account of certain further observations.

The methods used to determine the antibody content of various fluids are the same as were used by Dr. Carlson and myself. The cerebrospinal fluids used in these experiments were obtained without any admixture whatsoever of blood, and they appeared absolutely clear. In no case were there any inflammatory or other changes in the membranes and other structures of the central nervous system.

1. Does the cerebrospinal fluid inhibit agglutination of rat corpuscles?

I have found that in mixtures of normal and immune dog serum and rat corpuscles, the replacement of the salt solution with cerebrospinal fluid of normal dogs in no way diminishes the agglutinative action of the serum.

Several dogs, given a single intravenous injection of rat blood, one cubic centimeter of a 10 per cent suspension per kilo of weight, have been killed at varying intervals and bled dry so that the cerebrospinal fluid could be withdrawn easily without the slightest admixture of blood. This quantity of rat blood injected intravenously causes an abundant production of antibodies and may be regarded as an optimum antigenic dose for the dog.

The serum of one such dog (223) killed on the eighth day after injection, agglutinated and opsonized rat corpuscles in a dilution of 1 to 768. The cerebrospinal fluid did not agglutinate the corpuscles but it had a distinct opsonic effect at 1 to 96. The addition of this fluid to the serum of this dog and to normal dog serum in place of salt solution in preparing dilutions of varying strengths, and allowing the mixture to remain at 37° C. for two to three hours, did not reduce the agglutinative power of the sera at all. The same results were obtained with the cerebrospinal fluids from other dogs (187, 232, 233) in both earlier and later stages of antibody formation, the opsonic value of the fluid varying from 1–24 to 1–384.

From these results it may be concluded that the cerebrospinal fluid of dogs, whether normal or immunized with rat blood, does not inhibit the agglutinative action of dog serum on rat corpuscles.

2. Is the opsonic action of the cerebrospinal fluid of dogs injected with rat blood due to the same substance as the opsonin in the blood and lymph?

The indications all point to the presence in the cerebrospinal fluid of the same opsonin as in the blood and lymph.

In the first place the concentration though much less than in the blood and lymph describes the same general course as shown in Chart 1. Other estimations of the opsonin content of the blood and cerebrospinal fluid of immunized dogs, the two estimations in each case being made under exactly comparable conditions, give the following results:

Blood	Cerebrospinal Fluid	Blood	Cerebrospinal Fluid
6,144	384	768	48
768	24	1,536	96
1,536	48	1,536	96

In every case the agglutinin content of the blood corresponded closely to the opsonin content while the cerebrospinal fluid was devoid of all agglutinative effect. Furthermore, the opsonin in the cerebrospinal fluid is specific for rat corpuscles to which it becomes bound so that the fluid loses opsonic power after being treated for an hour or two with a sufficient amount of corpuscles and then centrifugated clear.

The opsonic power of the fresh fluid of dogs injected with rat corpuscles does not seem to be diminished by heating the fluid to 58° C. for 30 minutes. Inasmuch as the opsonic power is intensified by the addition of normal dog serum in quantities so small as to have no demonstrable opsonic action by themselves (Table 1), it may be concluded that the fluid contains a specific, thermostable opsonin which cannot be differentiated from that in the immune serum, that is, in the blood and lymph.

TABLE 1.

REACTIVATION OF THE OPSONIN IN CEREBROSPINAL FLUID.

Cerebrospinal Fluid of Immunized Dog	Fresh Dog Serum	Phagocytosis of Rat Corpuscles
.025 c.c.	.....	0
.025 c.c.	.005 c.c.	+
.025 c.c.	.0025 c.c.	+
.....	.005 c.c.	0

3. Are lysin, precipitin, and complement-deviating antibodies present in the cerebrospinal fluid of dogs injected with rat blood?

I have not been able to obtain any evidence that the cerebrospinal fluid of dogs immunized with rat blood contains any lysin for rat corpuscles. Lysis does not occur in mixtures of rat corpuscles and cerebrospinal fluid to which complement is added in the form of guinea-pig serum. As rat corpuscles do not seem to be laked easily by dog lysin it is of course possible that the fluid may contain lysin in quantities so small as to escape detection by the usual method.

Neither has it been possible to detect the presence of any complement binding substances in the cerebrospinal fluid of dogs injected with rat blood and the serum of which, withdrawn at the same time as the fluids, readily bound the complement in guinea-pig serum in mixtures with rat corpuscles. The indicator used was antisheep rabbit serum and sheep corpuscles. The results obtained are illustrated by the experiment given in Table 2.

Finally, precipitin for rat serum has not been demonstrated in the cerebrospinal fluid even when the corresponding dog serum (262) caused a precipitate to form in rat serum diluted 1:1280.

TABLE 2.

EXPERIMENT ON COMPLEMENT-DEVIATION BY SERUM AND CEREBROSPINAL FLUID OF DOG INJECTED WITH RAT BLOOD.

ANTIGEN (5 PER CENT RAT BLOOD)	SERUM OR CEREBROSPINAL FLUID OF DOG INJECTED WITH RAT BLOOD	COMPLEMENT (GUINEA-PIG SERUM)	HEMOLYTIC SYSTEM		LYSIS
			Antisheep Amboceptor	Sheep Corpuscles 5 Per Cent	
0.2 c.c.	Serum 0.05 c.c.	0.0125 c.c.	0.007 c.c.	0.5 c.c.	0
"	" 0.003 "	"	"	"	0
"	Fluid 0.15 "	"	"	"	+++
"	" 0.1 "	"	"	"	+++
"	.....	"	"	"	+++
.....	Serum 0.5 "	"	"	"	+++

The serum agglutinated and opsonized rat corpuscles at a dilution of 1:1536.
The cerebrospinal fluid opsonized rat corpuscles at a dilution of 1:96.

4. Does opsonin enter the cerebrospinal fluid after passive immunization with respect to rat blood?

So far as the antibodies for goat blood are concerned Dr. Carlson and I found the distribution after passive immunization by transfusion to correspond closely to the distribution in actively immunized animals. We made no observations, however, on the distribution of antibodies for rat corpuscles after passive immunization. I have found that in dogs injected with antirat dog serum the cerebrospinal fluid acquires some opsonic but no agglutinative power with respect to rat corpuscles. The observations in one case were made twenty-four hours after the intracardiac injection of the immune serum, and at this time the serum of the injected animal agglutinated rat corpuscles in a dilution of 1:768 and opsonified at 1:384, while the cerebrospinal fluid was opsonic at 1:6.

SUMMARY.

It appears that the cerebrospinal fluid of the dog does not inhibit the agglutination of rat corpuscles by the serum of this animal; that the opsonic effect of cerebrospinal fluid of dogs injected with rat blood is due to a specific, thermostable opsonic substance, seemingly identical with that in the blood and lymph, this being the only antibody demonstrable in the cerebrospinal fluid of dogs so immunized; and finally that this opsonin makes its way into the fluid after passive immunization.

These results indicate that opsonin can occur as a distinct substance and that its presence in the cerebrospinal fluid is the outcome of some form of selective process.